IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:	: MORSEMAN, et al.	Confirmation No.: 6731
Application No.:	: 09/882,376	
Filed	: June 18, 2001	
Title	: HIGH FLUORESCENT INTENSITY CROSS-LINKED ALLOPHYCOCYANIN	
TC/Art Unit	: 1641	
Examiner:	: Counts, Gary W.	
Docket No.	: 62611.000202	
Customer No.	: **21967**	

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 C.F.R. § 1.132

Sir:

I, Mark Wesley Moss, declare that:

1.) I attended The Johns Hopkins University from 1988 through 1992, in pursuit of a Bachelor of Science degree in Chemical Engineering.

2.) I have been employed by Martek Biosciences Corporation ("Martek") since 1997, where I have performed research in the field of immunoassay design and flow cytometry, and have co-authored several peer reviewed publications relating to the use of algal phycobiliproteins in various immunoassay applications, including flow cytometry. A recitation of some of these publications, together with details of my education, are given in the short version of my curriculum vitae, which is attached as **Exhibit A**.

3.) While employed at Martek, I participated in the development of a new method for stabilizing allophycocyanin ("APC") which avoided the use of strongly chaotropic agents during cross-linking and recovery of APC. The resultant cross-linked APC (hereinafter "SL-APC")

which had not been exposed to strongly chaotropic agents was offered for sale to a third party as described in my previous declaration, which was submitted to the U.S. Patent and Trademark Office on September 29, 2005.

4.) Subsequently, it was discovered that the use of SL-APC in time-resolved fluorescence assays conferred enhanced performance compared to conventional cross-linked APC (hereinafter "XL-APC"), and the patent application designated by U.S. Serial No. 09/882,376 ("the '376 application") was prepared and filed. I am a named inventor of the '376 application.

5.) I have also performed, supervised, and monitored quality control ("QC") assays for SureLight®–APC-Streptavidin ("SLAPC-SA"), which is prepared according to the process described in the '376 application and offered for sale by Martek, for a period of six years.

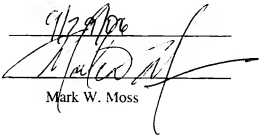
6.) QC assays of SLAPC-SA, which test its activity relative to the standard crosslinked-APC-Streptavidin ("XLAPC-SA") in time-resolved fluorescence assays, have been performed on 43 different lots of SL-APC. The streptavidin component of the conjugates bind to a biotinylated tyrosine-containing peptide. In the positive control wells of the QC assays, a fraction of tyrosine-containing peptides are phosphorylated, which allows them to be bound by a Europium-labeled anti-phosphotyrosine specific antibody ("anti-pTyr-Eu"). In the negative control wells of the QC assays, all of the tyrosine-containing peptides are unphosphorylated and therefore, will not be bound by a anti-pTyr-Eu. Whenever a sample well contains phosphorylated peptides, the SLAPC-SA (or XLAPC-SA) and anti-pTyr-Eu can bind to the same peptide and are brought into proximity with each other. The well is then excited with 340 nm light, which is absorbed by the bound Eu component, and energy is transferred by a non-radiative, resonant means to the bound SLAPC (or XLAPC) component, which emits light at 665 nm. When no phosphorylated peptides are present, the Eu and SLAPC (or XLAPC) do not come into proximity to each other and no energy is transferred; therefore, very little signal is produced at 665 nm. The ratio of signals from the positive and negative wells are calculated and the ratios for the SLAPC-SA samples are compared to the XLAPC-SA control ratios, which are used as a benchmark. Statistical analysis of the QC assays, as shown in **Exhibit B**, indicates that the

signal-to-noise ratios of SLAPC-SA compared to XLAPC-SA are, in fact, statistically significant ($p < 0.05$).

7.) Although the resulting signal-to-noise ratios of the QC assays are higher than the signal-to-noise ratios in Example 6 of the '376 application, the QC assays were performed using the SL-APC prepared by the method described in the '376 application and used in Example 6. Example 6, however, used src-tyrosine kinase to phosphorylate all of the peptides. Therefore, the amount of tyrosine that was phosphorylated on each peptide used in Example 6 was less than the amount of tyrosine that was phosphorylated on each of the phosphorylated peptides used in the QC assay. Because the phosphorylation methods used in the two assays are different, it would be reasonable to obtain a different magnitude of signal-to-noise ratios. For example, the increased amount of phosphorylated tyrosine on each peptide used in the QC assay may cause the emission of more light, thereby causing a greater signal, because more SLAPC-SA (or XLAPC-SA) and anti-pTyr-Eu are in close proximity to each other.

8.) The undersigned acknowledges that willful false statements and the like are punishable by fine or imprisonment, or both (18 U.S.C. § 1001), and may jeopardize the validity of the application or any patent issuing thereon. The undersigned declares further that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true.

Executed on: 9/27/06

Declarant's Signature: 

Mark W. Moss

EXHIBIT A

CURRICULUM VITAE
Mark Wesley Moss

Current Position

Martek Biosciences, Columbia, MD- Group Leader, Research and Development, Detection Systems Group

Professional Background

2002-04 Martek Biosciences, Columbia, MD- Research Associate III, Research and Development, Detection Systems Group

1997-97 Martek Biosciences, Columbia, MD- Research Associate II, Fluorescent Products Group

1994-97 The Johns Hopkins University School of Medicine, Division of Infectious Disease, Baltimore, MD- Senior Laboratory Technician II, International HIV/AIDS Reference Laboratory, NIH/NIAID

1990-94 The Johns Hopkins University School of Medicine, Division of Infectious Disease, Baltimore, MD- Laboratory Technician, International HIV/AIDS Reference Laboratory, NIH/NIAID

Education

1988-92 The Johns Hopkins University, Baltimore MD. – BS Program, Chemical Engineering

Societies

The Society for Biomolecular Screening
The International Society for Analytical Cytology
The Chesapeake Cytometry Consortium

Professional Experience

Laboratory

- Protein microarray design and optimization
- Flow cytometry, including operation and maintenance of in-house fluorescent cell sorter
- Isolation, derivitization, and bioconjugation of fluorescent proteins and other biomolecules
- Fluorescent, chemifluorescent, and enzyme-linked immunoassays and assay development
- UV/Vis absorbance and fluorescence emission spectroscopy
- Aqueous and organic chemistry methods
- Western blotting
- Mammalian cell culture
- Column chromatography
- PCR and RT-PCR

Business development

- Intellectual property development
- Tradeshows and conferences
- Conversion of data and technical works into marketing materials
- Graphics for tradeshows and marketing pieces

Other Experience

Biosafety level 3 training and work experience
Yearly OSHA Hazards Communications training

Publications

Telford WG. Moss MW. Morseman JP. Allnutt FCT. Cyanobacterial Stabilized Phycobilisomes as Fluorochromes for Extracellular Antigen Detection by Flow Cytometry. JOURNAL OF IMMUNOLOGICAL METHODS. 254(1-2): 13-30, 2001 Aug 1.

Telford WG. Moss MW. Morseman JP. Allnutt FCT. Cryptomonad Algal Phycobiliproteins as Fluorochromes for Extracellular and Intracellular Antigen Detection by Flow Cytometry. CYTOMETRY. 44(1): 16-23, 2001 May 1.

Zoha SJ. Ramnarain S. Morseman JP. Moss MW. Allnutt FCT. Rogers Y. Harvey B. PBXL Fluorescent Dyes for Ultrasensitive Direct Detection. JOURNAL OF FLUORESCENCE. 9(3):197-208, 1999 Oct.

Morseman JP. Moss MW. Zoha SJ. Allnutt FC. PBXL-1: a new fluorochrome applied to detection of proteins on membranes. BIOTECHNIQUES. 26:559-63, 1999 Mar.

Moss MW. Carella AV. Provost V. Quinn TC. Comparison of absolute CD4+ lymphocyte counts by enzyme immunoassay(TRAx CD4 Test Kit) and flow cytometry. CLINICAL AND DIAGNOSTIC LABORATORY IMMUNOLOGY. 3(4):371-3, 1996 Jul.

Carella AV. Moss MW. Provost V. Quinn TC. A manual bead assay for the determination of absolute CD4+ and CD8+ counts in human immunodeficiency virus-infected individuals. CLINICAL AND DIAGNOSITIC LABORATORY IMMUNOLOGY. 2(5):623-5, 1995 Sep.

Asihene PJ. Kline RL. Moss MW. Carella AV. Quinn TC. Evaluation of rapid test for detection of antibody to human immunodeficiency virus type 1 and type 2. JOURNAL OF CLINICAL MICROBIOLOGY. 32(5):1341-2, 1994 May.

Winsom C. DePaola LG. Thomas DL. Moss MW. Meiller J. Overholser C. Quinn TC. Prevalence of hepatitis B virus (HBV) and hepatitis C virus (HCV) in dental school patients and dental practitioners in Maryland. JOURNAL OF THE MARYLAND STATE DENTAL ASSOCIATION. 1993 Spring;35(1):17-9.

Quinn TC. KlineRL. Moss MW. Livingston RA. Hutton N. Acid dissociation of immune complexes improve diagnostic utility of p24 antigen detection in perinatally acquired human immunodeficiency virus infection. JOURNAL OF INFECTIOUS DISEASES. 167(5):1193-6, 1993 May.

Bollinger RC Jr. Kline RL. Francis HL. Moss MW. Bartlett JG. Quinn TC. Acid dissociation increases the sensitivity of p24 antigen detection for the evaluation of antiviral therapy and disease progression in asymptomatic human immunodeficiency virus-infected persons. JOURNAL OF INFECTIOUS DISEASES. 165(5):913-6, 1992 May.

Abstracts

Johnson T. Morseman J. Ramnarain S. Zoha S. Moss M. Allnutt FCT. Direct fluorescent detection using PBXL dyes. IBC CONFERENCE ASSAY MINIATURIZATION, SQUAW VALLEY, NEVADA FEBRURARY 1998.

Kline R. Newhouse R. Granade T. Phillips S. Moss M. Quinn TC. Evaluation of the MicroTrak II HIV-1/HIV-2 Recombinant Antigen Enzyme Immunoassay. 94th GENERAL MEETING OF THE AMERICAN SOCIETY OF MICROBIOLOGY, LAS VEGAS, NEVADA MAY 1995.

Kelen GD. Hexter DA. Kline R. Carella A. Moss M. Quinn TC. Dynamics of the HIV epidemic in an inner-city emergency department (ED): implications for ED based targeted screening. IX INTERNATIONAL CONFERENCE ON AIDS, BERLIN, GERMANY JUNE 1993.

Quinn TC. Kline R. Livingston R. Carella A. Moss M. Hutton N. An algorithm for the early diagnosis of perinatally acquired HIV-1 infection using the IgA immunoblot assay and the modified p24 antigen (Ag) assay. IX INTERNATIONAL CONFERENCE ON AIDS, BERLIN, GERMANY JUNE 1993.

Presentations

Comparison of absolute $CD4^+$ lymphocyte counts by enzyme immunoassay(TRAx CD4 Test Kit) and flow cytometry. Oral presentation, Emerging Technologies Satellite Symposium- IX INTERNATIONAL CONFERENCE ON AIDS, BERLIN, GERMANY JUNE 1993.

Patents

Morseman JP. Moss MW. Allnutt FCT. High Fluorescent Intensity Crosslinked Allophycocyanin. [pending].

Morseman, JP. Moss MW. Ellis LA. Reelin Deficiency or Dysfunction and Methods Related Thereto. [pending]

EXHIBIT B

s/n = signal at 665 nm from positive (hot) wells/ signal at 665 nm from negative (cold) wells

	Martek s/n	Competitor s/n
Lot 1	19.32	5.08
Lot 2	13.02	4.49
Lot 3	19.01	4.06
Lot 4	25.46	5.17
Lot 5	20.41	3.80
Lot 6	20.49	9.60
Lot 7	19.48	10.41
Lot 8	27.23	14.09
Lot 9	19.84	9.87
Lot 10	20.67	11.13
Lot 11	21.95	9.59
Lot 12	19.64	8.75
Lot 13	23.84	10.16
Lot 14	22.60	9.11
Lot 15	25.61	8.36
Lot 16	23.00	6.79
Lot 17	16.70	5.02
Lot 18	20.99	6.04
Lot 19	25.33	7.07
Lot 20	21.18	4.73
Lot 21	21.35	4.73
Lot 22	23.44	5.84
Lot 23	23.39	5.64
Lot 24	22.16	5.09
Lot 25	25.32	7.11
Lot 26	7.37	4.63
Lot 27	16.09	5.88
Lot 28	21.40	7.31
Lot 29	19.28	6.59
Lot 30	20.85	6.55
Lot 31	17.41	7.78
Lot 32	16.36	7.01
Lot 33	16.12	7.86
Lot 34	16.84	7.46
Lot 35	12.43	5.53
Lot 36	13.03	6.07
Lot 37	19.30	6.05
Lot 38	31.28	11.94
Lot 39	24.91	7.32
Lot 40	28.93	8.92
Lot 41	17.67	7.41
Lot 42	21.92	7.64
Lot 43	25.35	6.91
Mean	20.65	7.22
StdDev	4.58	2.26
CV	0.22	0.31

Parameter	Value
Table Analyzed	Data 2
Column A	martek s/n
vs	vs
Column B	competitor s/n
Paired t test	
P value	P<0.0001
P value summary	***
Are means signif. different? (P < 0.05)	Yes
One- or two-tailed P value?	Two-tailed
t, df	t=21.5145 df=42
Number of pairs	43
How big is the difference?	
Mean of differences	13.4274
95% confidence interval	12.1674 to 14.6875
R squared	0.916811
How effective was the pairing?	
Correlation coefficient (r)	0.449145
P Value (one tailed)	0.0013
P value summary	**
Was the pairing significantly effective?	Yes

	Martek s/n	Competitor s/n
Number of values	43	43
Minimum	7.37	3.8
25% Percentile	17.67	5.53
Median	20.85	7.01
75% Percentile	23.44	8.75
Maximum	31.28	14.09
Mean	20.6505	7.22302
Std. Deviation	4.57507	2.25592
Std. Error	0.697692	0.344024
Lower 95% CI	19.2425	6.52875
Upper 95% CI	22.0585	7.9173